REMARKS

Claim 1 has been amended to specify that the medium comprises a selection agent in addition to the differentiation agent. Claim 1 has also been amended to change "osmoticum" to "polyethylene glycol" and to specify that the amount of gelling agent present is either between 3% to 5% or between 0.5% and 1.5%. Support for these amendments can be found in the claims as originally filed. Claim 1 has further been amended to include a step of regenerating plants from the selected pine cells. Support for this amendment can be found at page 15, lines 3-9 of the specification. Claims 3, 5, 6, 7, 10, 14-16, 19, 22, 28, 29, 31 and 37 have been amended to be consistent with the amendments to claim 1.

Claims 44-55 have been canceled without prejudice to filing in a continuation application.

It is submitted that these amendments do not constitute new matter and their entry is requested.

Applicants would like to thank the Examiner and his supervisor for the courtesies extended to the undersigned and the inventor, Dr. Marie Connett-Porceddu, at the interview on 26 June 2003. During the interview Dr. Connett-Porceddu described the present invention in the context of the differences between hard and soft pines and the prior inability to recover selected cells at a level which would be commercially useful for producing transgenic pine, especially for Southern yellow pines and hybrids thereof, which are hard pines. According to Dr. Connett-Porceddu, one feature of the invention which enabled the recovery of transgenic cells for regenerating transgenic plants was the use of a differentiation agent in the selection medium. The use of the differentiation agent was in addition to the selection agent which is used to select transgenic cells. The use of the differentiation agent ensured that transgenic cells could be recovered following selection and these transgenic cells could be regenerated into transgenic plants in commercially useful quantities. For example, prior art techniques resulted in a 26% rate of regeneration for loblolly pine (*Pinus taeda*), whereas the present invention resulted in a 71% rate of regeneration. Similarly, for elite families, the prior art was unable to achieve regeneration, whereas the present invention achieved an 80% rate of regeneration. The differences between hard and soft pines and the unobvious nature of plant

transformation and regeneration in these species was discussed. The Examiner suggested that Declarations be submitted to address this point as well as the success of the present invention. Applicants are in the process of preparing Rule 132 Declarations, and these will be submitted as soon as they have been executed and received by the undersigned.

The Examiner rejected claims 44-55 under 35 U.S.C. § 112, first paragraph for lack of written description. Although Applicants do not agree with this rejection, Applicants have canceled these claims in an effort to expedite the prosecution of the remaining claims without prejudice to filing in a continuation application.

The Examiner rejected claims 1-3, 22-23, 28, 31-32, 37, 40 and 42 under 35 U.S.C. § 102(b) as being anticipated by Wenck et al. (*Plant Molecular Biology* 39:407-416, 1999). Wenck et al. does describe the use a differentiation agent selected from the group consisting of ABA, PEG or a gelling agent in the specified concentration in a selection medium which is used for selecting transgenic embryogenic pine cells of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Thus, Wenck et al. cannot anticipate the claims.

Furthermore, Applicants note that the claims are directed to the regeneration of genetically modified plants of pine of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Wenck et al. teaches the regeneration of genetically modified plants of Norway spruce (*Picea abies*). Wenck et al. does not teach the regeneration of genetically modified plants of Southern yellow pines, such as *Pinus taeda*, and hybrids thereof. In fact, Wenck et al. specifically teaches that it was unable to recover stable transformants through selection for loblolly pine (i.e., *Pinus taeda*). See page 413, left column. Since Wenck et al. did not recover stable transformants and did not regenerate genetically modified plants of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof, it cannot anticipate the presently claimed invention.

For these reasons, it is submitted that Wenck et al. does not anticipate the claimed subject matter, and withdrawal of this rejection is requested.

The Examiner has rejected claims 1-3, 5-8, 10, 11, 14-17, 19, 20, 22, 23, 28, 29, 31, 32, 37, 38, 40 and 42 under 35 U.S.C. §103(a) as being obvious over Wenck et al. taken with Rutter et al. (US 5,731,204). As shown above, Wenck et al. does not disclose the elements of the claimed invention, i.e., the use of the claimed differentiation agents in a selection medium. These elements are not supplied by the secondary reference. Therefore, the cited references cannot render the claimed invention obvious.

Specifically, there is no disclosure of the transformation of pine of the genus *Pinus* in Rutter et al., and thus, no disclosure or enhancing transformation efficiency. Since there is no disclosure of transformation, there is no disclosure of selection of transgenic embryogenic cells. Thus, Rutter et al. does not disclose the use of ABA, PEG or a gelling agent in the specified amount in a selection medium which is used for selecting transgenic embryogenic pine cells of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Furthermore, since Rutter et al. does not relate to transformation, there is no motivation to combine this reference with Wenck et al. For these reasons, it is submitted that Wenck et al. taken with Rutter et al. does not render the claimed subject matter obvious, and withdrawal of this rejection is requested.

The Examiner rejected claims 1-3, 22-28, 31-37 and 40-43 under 35 U.S.C. § 103(a) as being unpatentable over Wenck et al. taken with Levee et al. (*Molecular Breeding* 5:429-440, 1999). As shown above, Wenck et al. does not disclose the elements of the claimed invention. These elements are not supplied by the secondary reference. Therefore, the cited references cannot render the claimed invention obvious.

Specifically, Levee et al. does not disclose the use of ABA, PEG or a gelling agent in the specified amount in a selection medium for selecting transgenic embryogenic pine cells of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Thus, the combination of Wenck et al. and Levee et al. does not render the claimed invention obvious.

Furthermore, Levee et al. discloses *Agrobacterium* transformation of white pine, *Pinus strobes*. As is well known in the art, white pine is a soft pine and not a hard pine, such as the Southern yellow pines and hybrids thereof. As is evident in the name, *Pinus strobus*, white pine is

a member of the subgenus *Strobus* and is not a member of the subgenus *Pinus*. For example, most classifications of *Pinus* recognize two major lineages: subgenus *Strobus* (haploxylon or soft pines, with one fibrovascular bundle in the needle) and subgenus *Pinus* (diploxylon or hard pines, with two fibrovascular bundles in the needle). This division is consistent with data from wood anatomy and secondary chemistry, and is supported in recent molecular phylogenetic studies (Strauss and Doerksen, 1990, *Evolution* **44**:1081-1096; Wang and Szmidt, 1993, *Plant Systematics and Evolution* **188**:197-211; reviewed in Price et al., 1998, in *Ecology and Biogeography of Pinus*, Cambridge University Press, Cambridge, pp. 49-68).

Pines have a relatively rich fossil record dating back to the Early Cretaceous, 130 million years ago (review in Axelrod et al., 1986, *Ann Mo Bot Gard* **73**:565-641; Klaus et al., 1989, *Plant Systematics and Evolution* **162**:133-163; Van der Burgh, 1973, *Review of Paleobotany and Palynology*, **15**:73-275; Millar, 1993, *Ann Mo Bot Gard* **80**:471-498). The genetic distance between subgenera, at least between *Pinus* and *Strobus*, may be as large as, or larger than the genetic distance between other conifer genera, e.g., between *Cedrus* and *Abies* (Price et al., 1987, *Systematic Botany*, **12**:91-97), and if strict genetic criteria were used, they should perhaps be treated at generic rank. As is commonly known, hard pines are unable to breed with soft pines, though they can interbreed readily, if the correct timing and other conditions are provided, with other hard pine species (a seminal reference is Critchfield and Little, 1966, *Geographic distribution of the pines of the world*, USDA Forest Service Miscellaneous Publication 991, Washington, D.C.; see also Little and Critchfield, 1969, *Subdivision of the genus Pinus pines*, USDA Forest Service Miscellaneous Publication 1144, Washington, D.C.). Hard pines are unaffected by a number of diseases, such as white pine blister rust, that readily infect soft pines. Their susceptibility to *Agrobacterium* infection appears to be quite different as well.

Levee et al. discloses the transformation and regeneration of pine of the subgenus *Strobus* which, according to this reference, "is the first work on genetic transformation on **this pine species** as well as the first report of successful stable genetic transformation of **a pine species** using a disarmed strain of *A. tumefaciens*". (See page 36, first paragraph of Discussion, emphasis added).

Levee et al. does not disclose the transformation and regeneration of pine of the subgenus *Pinus*. The amended claims are clearly directed to pine cells of the *Pinus* genus. It is well known to those skilled in the art that somatic embryogenesis systems for soft pines are different from those for hard pines, such as Southern yellow pines and hybrids thereof. It is not insignificant that Levee et al. utilized a soft pine which is more easily regenerated than hard pines. Although the Examiner cited art showing transformation and regeneration of soft pine, he has not cited any art showing transformation and regeneration of hard pines as set forth in the claims. Furthermore, it is submitted that there have been no reports in the literature of the regeneration of plants following stable transformation of embryogenic cultures of any pines of the *Pinus* subgenus by *Agrobacterium*. In order to further establish these known differences between hard and soft pines and to provide further evidence of the nonobviousness of the present invention, Applicants are in the process of preparing Rule 132 Declarations and these will be submitted as soon as they have been executed and received by the undersigned.

For these reasons, it is submitted that Wenck et al. and Levee et al. do not render the claimed subject matter obvious, and withdrawal of this rejection is requested.

The Examiner rejected claims 1-3, 22-28, 30-37 and 39-43 under 35 U.S.C. § 103(a) as being unpatentable over Wenck et al. taken with Levee et al. and Rutter et al. As shown above, Wenck et al. does not disclose the elements of the claimed invention and these elements are not supplied by the secondary references. Therefore, the cited references cannot render the claimed invention obvious.

Specifically, none of the cited references disclose the use of ABA, PEG or a gelling agent in the specified amount in a selection medium which is used for selecting transgenic embryogenic pine cells of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Furthermore, none of the cited references disclose the regeneration of genetically modified plants of the genus *Pinus* selected from the group consisting of Southern yellow pines and hybrids thereof. Thus, the combination of Wenck et al., Levee et al. and Rutter et al. does not render the claimed invention obvious.

The Examiner provisionally rejected claims 1-3, 5-8, 10, 11, 14-17, 19, 20 and 22-55 under the judicially created doctrine of obviousness-type double patenting over claims 1, 12-14, 15, 17, 18, 21, 23, 25, 30, 34, 45, 47, 51, 57 and 63-81 of U.S. patent application Serial No. 09/973,088 (the '088 application). It is submitted that the Examiner is in error in this rejection.

Specifically, the claims of the present invention are directed to a method for regenerating genetically modified plants of Southern yellow pines and hybrids thereof. One step of the claimed method is the selection of transgenic embryogenic pine cells using a selection medium comprising a selection agent and an agent that regulates differentiation of embryos from embryogenic cells. The differentiation agent is selected from the group consisting of ABA, PEG and a gelling agent of the specified amount. This step is neither described nor claimed in the '088 application. There is nothing in the '088 application which would suggest using a differentiation agent in the medium for selection of transgenic embryogenic pine cells, nor has the Examiner pointed to any teaching in the '088 application for the use of the claimed differentiation agents in a selection medium. Therefore, it is submitted that the present claims are not unpatentable for obviousness-type double patenting over the cited claims of the '088 application. Withdrawal of this rejection is requested.

In view of the above amendments and remarks, in conjunction with the remarks made in the previous amendment, it is believed that the claims satisfy the requirements of the patent statutes and are patentable over the prior art. Reconsideration of the instant application and early notice of

allowance are requested. The Examiner is invited to telephone the undersigned if it is deemed to expedite allowance of the application.

Respectfully submitted,

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